

O'Bryen, Barbara

From: Chan, Christina
Sent: Friday, March 23, 2001 9:04 AM
To: Bui, Phuong; O'Bryen, Barbara
Subject: RE: 09/410835 sequence search
Importance: High

Please rush. Thanks Chris

Chris Chan.
TC 1600 New Hire Training Coordinator and SPE, 1644
CM 1, Room 9B19
308-3973

-----Original Message-----

From: Bui Phuong
Sent: Thursday, March 22, 2001 7:39 PM
To: Chan, Christina
Subject: 09/410835 sequence search

Chris,

Please forward the following search to **Barb O'Bryen** as a rush. 2-month amendment. Thanks. P.

Applic. No. 09/410835
Phuong Bui
AU 1638
CM1, 9A09; Mailbox 9E12
305-1996

Please search 15-mers of SEQ ID NO. 4. Since this case has priority back to 1992, please list citations for the top 200 hits.

Please include an interference search of SEQ ID NO:4.

Thank you! Phuong.

POINT OF CONTACT:
BARB O'BRYEN
TECH. INFORMATION SPECIALIST
STIC CM1 12C14 308-4291

BoB
3-27-01

L15 ANSWER 188 OF 209 MEDLINE
 AN 87246068 MEDLINE
 DN 87246068 PubMed ID: 3109864
 TI A genetically engineered P450 monooxygenase: construction of the functional fused enzyme between rat cytochrome P450c and NADPH-cytochrome P450 reductase.
 AU Murakami H; Yabusaki Y; Sakaki T; Shibata M; Ohkawa H
 SO DNA, (1987 Jun) 6 (3) 189-97.
 Journal code: EAW; 8302432. ISSN: 0198-0238.
 CY United States
 DT Journal; Article; (JOURNAL ARTICLE)
 LA English
 FS Priority Journals
 EM 198708
 ED Entered STN: 19900305
 Last Updated on STN: 19970203
 Entered Medline: 19870810
 AB A hybrid cDNA encoding a fused enzyme consisting of rat cytochrome P450c and rat NADPH-cytochrome P450 reductase was constructed by combining the cytochrome P450c cDNA with the cDNA fragment encoding the protease-solubilized moiety of the NADPH-cytochrome P450 reductase. The hybrid cDNA was inserted between the yeast alcohol dehydrogenase I promoter and terminator of the expression vector pAAH5 to yield expression plasmid pAMP19. *Saccharomyces cerevisiae* AH22 cells transformed with the expression plasmid pAMP19 produced a **130-kD** protein reactive with both anti-cytochrome P450c Ig and antireductase Ig. The yeast cells containing the fused enzyme exhibited about four times higher monooxygenase activity toward 7-ethoxycoumarin than those containing rat cytochrome P450c alone. The fused enzyme was purified from the yeast microsomal fraction by sequential chromatography with DEAE-cellulose and 2',5'-ADP Sepharose 4B columns. The preparation had an apparent molecular weight of **130 kD** and the same sequence of the 10 amino-terminal amino acids as that of rat cytochrome P450c. Spectral properties of the fused enzyme indicated the presence of a protoheme, flavin adenine dinucleotide, and flavin mononucleotide in the molecule. The reaction mechanism of the fused enzyme followed first-order kinetics. These results clearly indicate that the fused enzyme is a new self-catalytic P450 monooxygenase. Trypsin treatment of yeast microsomes containing the fused enzyme suggested that the P450 moiety is embedded in the microsomal membrane with the reductase moiety lying on the cytoplasmic

L15 ANSWER 208 OF 209 MEDLINE
 AN 85150284 MEDLINE
 DN 85150284 PubMed ID: 6241481
 TI Cloning and expression in Escherichia coli of the TL-DNA gene 4 of Agrobacterium tumefaciens under the control of the PR promoter of bacteriophage lambda.
 AU Sibold L; Guiso N; de Beuckeleer M; Van Montagu M
 SO BIOCHIMIE, (1984 Jul-Aug) 66 (7-8) 547-56.
 Journal code: A14; 1264604. ISSN: 0300-9084.
 CY France
 DT Journal; Article; (JOURNAL ARTICLE)
 LA English
 FS Priority Journals
 EM 198504
 ED Entered STN: 19900320
 Last Updated on STN: 19900320
 Entered Medline: 19850426
 AB A plasmid was constructed that directs expression of the TL-DNA gene 4 protein in E. coli. The different steps of the construction were as follows: i) a region of gene 4 encoding the amino-terminal portion of the protein was fused in frame to DNA encoding an enzymatically active carboxy-terminal fragment of beta-galactosidase. The hybrid gene was poorly expressed from the upstream lambda PL promoter carried by the vector. ii) in order to generate an efficient procaryotic ribosome binding site, a DNA fragment carrying the lambda PR promoter with the nearby Shine-Dalgarno (SD) sequence of gene cro was placed in front of the gene 4-lacZ fusion. A **recombinant** plasmid, termed pGV793, that expressed efficiently a fused protein 4-beta-galactosidase was identified among the Lac+ clones. DNA sequencing analysis showed that pGV793 carried a hybrid ribosome binding site composed of the cro SD' sequence, a five bp sequence and the ATG codon of gene 4. Plasmid pGV793 directed the synthesis of three polypeptides of molecular weight **132 Kd**, 126 Kd and 122 Kd that carried beta-galactosidase antigenic determinants. The largest polypeptide had the expected size for the hybrid protein. The fusion proteins which accounted for about 0.5% of the total cellular proteins were purified by immunoabsorption using anti-beta-galactosidase antiserum. iii) the complete gene 4 coding sequence was reconstituted, with the lambda PR promoter in place. The resulting pGV822 plasmid expressed a polypeptide whose molecular weight 27 Kd corresponded to the expected size for the gene 4 product. The pI was

6/7/5 (Item 5 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
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08404089 BIOSIS NO.: 000094121743
SERUM NEUTRALIZING ANTIBODY RESPONSE TO THE VACUOLATING **CYTOTOXIN** OF
HELICOBACTER-PYLORI
AUTHOR: COVER T L; CAO P; MURTHY U K; SIPPLE M S; BLASER M J
AUTHOR ADDRESS: DIV. INFECTIOUS DISEASES, A-3310 MEDICAL CENTER NORTH,
VANDERBILT UNIVERSITY SCH. MED., NASHVILLE, TENN. 37232-2605.
JOURNAL: J CLIN INVEST 90 (3). 1992. 913-918.
FULL JOURNAL NAME: Journal of Clinical Investigation
CODEN: JCINA
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: Approximately 50% of **Helicobacter pylori** isolates produce a **cytotoxin** in vitro that induces vacuolation of eukaryotic cells. To determine the in vivo relevance of this phenomenon, we sought to detect **cytotoxin**-neutralizing antibodies in sera from H. pylori-infected persons. As a group, sera from 29 H. pylori-infected patients neutralized the activity of the purified **cytotoxin** to a significantly greater extent than sera from 24 uninfected persons ($P = 0.007$). The **cytotoxin** neutralizing activity in sera from H. pylori-infected persons was mediated predominantly by the purified IgG fraction. Sera from H. pylori-infected persons neutralized the **cytotoxins** produced by multiple H. pylori strains, but failed to neutralize trimethylamine-induced cell vacuolation. Neutralization of **cytotoxin** activity by human or immune rabbit sera was associated with immunoblot IgG recognition of an 87-kD H. pylori protein. Similarly, neutralization of the toxin by sera was associated with IgG recognition of the purified **cytotoxin** in an enzyme-linked immunosorbent assay ($P < 0.0001$). The presence of **cytotoxin**-neutralizing antibodies in sera from H. pylori-infected persons indicates that the **cytotoxins** is synthesized in vivo.

? ds

Set	Items	Description
S1	1318984	PY=1992
S2	57204	HELICOBACTER OR CAMPYLOBACTER
S3	2672	S1 AND S2
S4	9988	CYTOTOXIN?
S5	41	S3 AND S4
S6	21	RD (unique items)
S7	1325243	PY=1991
S8	39882	PYLORI
S9	26	S7 AND S8 AND S2 AND S4
S10	15	RD (unique items)

6/7/6 (Item 6 from file: 5)
DIALOG(R)File 5: BIOSIS Previews(R)
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08378894 BIOSIS NO.: 000094109398
EXPRESSION OF 120 KILODALTON PROTEIN AND CYTOTOXICITY IN **HELICOBACTER**
-PYLORI
AUTHOR: CRABTREE J E; FIGURA N; TAYLOR J D; BUGNOLI M; ARMELLINI D;
TOMPKINS D S
AUTHOR ADDRESS: DEP. MED., ST. JAMES'S UNIV. HOSP., LEEDS LS9 7TF.
JOURNAL: J CLIN PATHOL (LOND) 45 (8). 1992. 733-734.
FULL JOURNAL NAME: Journal of Clinical Pathology (London)
CODEN: JCPAA
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: Antral biopsy culture supernatants from 14 subjects with chronic gastritis, known to have IgA antibodies to the 120 kilodalton protein, showed positive recognition of this antigen in western blots against a **cytotoxin** positive strain of **Helicobacter pylori** but gave negative reactions with two **cytotoxin** negative strains. Control immunoblots with culture supernatants from 13 non-responders to the protein were all negative. This indicates a direct association between expression of the 120 kilodalton protein in H. pylori strains and cytotoxicity.

JP

L15 ANSWER 161 OF 209 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 74
 AN 1988:242952 BIOSIS
 DN BA85:121354
 TI HIGH EXPRESSION OF FUNCTIONAL ADENOVIRUS DNA POLYMERASE AND PRECURSOR
 TERMINAL PROTEIN USING **RECOMBINANT** VACCINIA VIRUS.
 AU STUNNENBERG H G; LANGE H; PHILIPSON L; VAN MILTENBURG R T; VAN DER VLIET
 P
 C
 CS LAB. PHYSIOLOGICAL CHEMISTRY, STATE UNIV. UTRECHT, VONDELLAAN 24A, 3521
 GG UTRECHT, NETHERLANDS.
 SO NUCLEIC ACIDS RES, (1988) 16 (6), 2431-2444.
 CODEN: NARHAD. ISSN: 0305-1048.
 FS BA; OLD
 LA English
 AB Initiation of Adenovirus (Ad) DNA replication occurs by a protein-priming
 mechanism in which the viral precursor terminal protein (pTP) and DNA
 polymerase (pol) as well as two nuclear DNA-binding proteins from
 uninfected HeLa cells are required. Biochemical studies on the pTP and
 DNA polymerase proteins separately have been hampered due to their low
 abundance and their presence as a pTP-pol complex in Ad infected cells.
 We have constructed a genomic sequence containing the large open reading
 frame from the Ad5 pol gene to which 9 basepairs from a putative exon
 were ligated. When inserted behind a modified late promoter of vaccinia virus
 the resulting **recombinant** virus produced enzymatically active
140 kDa Ad DNA polymerase. The same strategy was applied
 to express the 80 kDa pTP gene in a functional form. Both proteins were
 overexpressed at least 30-fold compared to extracts from Adenovirus
 infected cells and, when combined, were fully active for initiation in an
 in vitro Adenovirus DNA replication system.

L15 ANSWER 172 OF 209 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 78
 AN 1988:375442 BIOSIS
 DN BA86:59352
 TI EIMERIA-ACERVULINA DNA CLONING AND CHARACTERIZATION OF **RECOMBINANT**
 SPOROZOITE AND MEROZOITE ANTIGENS.
 AU JENKINS M C; LILLEHOJ H S; DAME J B
 CS UNIV. FLORIDA, DEP. INFECTIOUS DISEASES, GAINESVILLE, FLORIDA 32610.
 SO EXP PARASITOL, (1988) 66 (1), 96-107.
 CODEN: EXPAAA. ISSN: 0014-4894.
 FS BA; OLD
 LA English
 AB Genes encoding antigens of Eimeria acervulina were cloned from cDNA
 expression libraries prepared from the sporozoite and merozoite stages in
 order to examine humoral and cellular immune responses to this protozoan
 parasite. Two clones expressing surface antigens were characterized by
 DNA hybridization studies to identify homologous genomic DNA fragments. The
 proteins they encode were identified by 125I-labeling, immunoblotting,
 immunofluorescence, and T-cell activation experiments. One, designated
 cSZ-1, encodes a **130-kDa** .beta.-galactosidase fusion
 protein which represents a portion of a p240/pl60 immunodominant
 sporozoite surface antigen. Immunofluorescence studies using anti-cSZ-1
 sera and live or 1% paraformaldehyde-fixed E. acervulina sporozoites have
 confirmed this surface locale. Purified cSZ-1 fusion protein, which is
 not recognized by sera from E. acervulina-infected chickens, induced the
 activation of immune T lymphocytes in vitro. Another cDNA clone,
 designated cMZ-8, gives rise to a **150-kDa** fusion
 protein and encodes a portion of a p250 immunodominant merozoite surface
 antigen. This was established by immunoblotting of 125I-labeled merzoite
 proteins with anti-cMZ-8 sera and immunofluorescence staining of live and
 1% paraformaldehyde-fixed E. acervulina merozoites. Purified cMZ-8 is
 recognized by sera from E. acervulina-infected chickens and induces a

L15 ANSWER 178 OF 209 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 81

AN 1987:490588 BIOSIS

DN BA84:125231

TI MAPPING OF THE GENE CODING FOR EPSTEIN-BARR VIRUS-DETERMINED NUCLEAR ANTIGEN EBNA3 AND ITS TRANSIENT OVEREXPRESSION IN A HUMAN CELL LINE BY USING AN ADENOVIRUS EXPRESSION VECTOR.

AU JOAB I; ROWE D T; BODESCOT M; NICOLAS J-C; FARRELL P J; PERRICAUDET M

CS INST. RECH. SCI. CANCER, CENT. NATL. RECH. SCI., 94800 VILLEJUIF, FR.

SO J VIROL, (1987) 61 (10), 3340-3344.

CODEN: JOVIAM. ISSN: 0022-538X.

FS BA; OLD

LA English

AB The open reading frame which lies within the Epstein-Barr virus (EBV) T2 cDNA isolated by Bodescot et al. (M. Bodescot, O. Brison, and M. Pericaudet, Nucleic Acids Res. 14:2611-2620, 1986) was inserted into a eucaryotic expression vector containing a strong adenovirus promoter. The T2 cDNA contains viral genomic sequences from the short BLRF3 open reading

frame fused to the adjacent BERF1 long open reading frame. After transfection of human cells, the **recombinant** plasmid directed the expression of a **140-kilodalton** protein. The expressed protein had the same molecular weight, subcellular

localization,

and immunological characteristics as the EBV-determined nuclear antigen EBNA3, which is made in lymphocytes latently infected with EBV.

Immunoprecipitation of extracts of transfected cells labeled with [32P]phosphoric acid showed that the EBNA3 protein is phosphorylated.

neutralizing antibodies to SA11 rotavirus.

L15 ANSWER 206 OF 209 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 98
AN 1986:358953 BIOSIS
DN BA82:63427
TI EXPRESSION OF MYCOPLASMA-PNEUMONIAE ANTIGENS IN ESCHERICHIA-COLI.
AU TREVINO L B; HALDENWANG W G; BASEMAN J B
CS UNIV. TEXAS HEALTH SCI. CENTER AT SAN ANTONIO, DEP. MICROBIOLOGY, SAN
ANTONIO, TEXAS 78284.
SO INFECT IMMUN, (1986) 53 (1), 129-134.
CODEN: INFIBR. ISSN: 0019-9567.
FS BA; OLD
LA English
AB A genomic library of Mycoplasma pneumoniae was generated by using
bacteriophage lambda EMBL3 as the vector. Screening of the library for
the expression of M. pneumoniae protein antigens with adsorbed anti-M.
pneumoniae serum revealed strong reactivity from a third of those clones
which contained mycoplasma DNA inserts. Three of the most highly reactive
clones were analyzed in detail and found to synthesize discrete
mycoplasma proteins. Two carried overlapping fragments of mycoplasma DNA which
encoded a protein that was readily detected in Escherichia coli after
infection with **recombinant** bacteriophage. The third clone
contained a novel mycoplasma DNA fragment which directed the synthesis of
two additional mycoplasma proteins. Further screening of the library with
antiserum raised against the major M. pneumoniae adhesin protein P1 (165
kilodaltons [kDa]) yielded one clone which produced an immunologically
reactive protein of **140 kDa**. Adsorption of anti-P1
serum by this clone selected a population of antibodies that were
reactive with M. pneumoniae adhesin P1 (165 kDa). These results
demonstrate that immunologically active M. pneumoniae proteins are

L15 ANSWER 196 OF 209 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 92
 AN 1986:338513 BIOSIS
 DN BA82:52717
 TI EXPRESSION OF THE HUMAN C-FMS PROTO-ONCOGENE PRODUCT COLONY-STIMULATING
 FACTOR-1 RECEPTOR ON PERIPHERAL BLOOD MONONUCLEAR CELLS AND
 CHORIOCARCINOMA CELL LINES.
 AU RETTENMIER C W; SACCA R; FURMAN W L; ROUSSEL M F; HOLT J T; NIENHUIS A W;
 STANLEY E R; SHERR C J
 CS DEP. OF TUMOR CELL, ST. JUDE CHILDREN'S RES. HOSP., MEMPHIS, TENN. 38105.
 SO J CLIN INVEST, (1986) 77 (6), 1740-1746.
 CODEN: JCINAO. ISSN: 0021-9738.
 FS BA; OLD
 LA English
 AB The c-fms gene product is related, and possibly identical, to the
 receptor
 the mononuclear phagocyte colony stimulating factor, CSF-1. Using
 antisera
 to a **recombinant** v-fms-coded polypeptide, glycoproteins encoded
 by the human c-fms locus were detected in mononuclear cells from normal
 peripheral blood and in promyelocytic HL-60 cells 24 h after induction of
 monocytic differentiation with phorbol ester. The **150-kD**
 human c-fms-coded glycoprotein was expressed at the cell surface, was
 active as a tyrosine-specific protein kinase in vitro, and shared
 primary
 structural features with the product of the feline retroviral v-fms
 oncogene. A biochemically indistinguishable glycoprotein was detected in
 human choriocarcinoma cells lines. Like peripheral blood mononuclear
 cells
 and phorbol ester-treated HL-60 cells, the choriocarcinoma cells
 expressed
 high affinity binding sites for human CSF-1. In addition to serving as a
 lineage specific growth factor in hematopoiesis, CSF-1, may play a role

7 2
Claim 38 (Amended). A [purified protein of the] recombinantly produced *Helicobacter pylori* cytotoxin (CT) polypeptide, wherein the recombinantly produced polypeptide exhibits substantially no toxicity, or substantially reduced toxicity.

Claim 39 (Amended). [The purified protein of claim 38 wherein said protein is] A recombinantly produced fragment of a *Helicobacter pylori* CT polypeptide, wherein the recombinantly produced fragment (i) comprises at least about ten amino acids, (ii) can induce the production of antibodies to *Helicobacter pylori*, and (iii) exhibits substantially no toxicity, or substantially reduced toxicity.

C 2
Claim 40 (Amended). A [polypeptide sequence of the *Helicobacter pylori* cytotoxin amino acid sequence set forth in] recombinantly produced *H. pylori* CT polypeptide or fragment thereof comprising SEQ ID NO:3 or a fragment thereof, which polypeptide [sequence] or fragment thereof : (i) comprises at least [five] about ten amino acids of SEQ ID NO:3, (ii) can induce the production of antibodies to *Helicobacter pylori*, and (iii) exhibits substantially no [contribution to] toxicity, or substantially reduced toxicity.

2 3
Claim 42 (Amended). The polypeptide [sequence] of claim 40, wherein said [sequence] polypeptide comprises [about five to] at least about fifteen amino acids.

C1-
Claim 43 (Amended). A prophylactic or therapeutic vaccine comprising an immunologically effective amount of a H. pylori CT polypeptide [sequence of the *Helicobacter pylori* cytotoxin amino acid sequence set forth in] comprising SEQ ID NO:3 or a fragment thereof, which polypeptide [sequence]: (i) comprises at least [five] about ten amino acids of SEQ ID NO:3, (ii) can induce the production of antibodies to *Helicobacter pylori*, and (iii) exhibits substantially no [contribution to] toxicity, or substantially reduced toxicity.

44. The vaccine of claim 43 wherein said sequence comprises at least ten amino acids.

Claim 45 (Amended). The vaccine of claim 43, wherein said [sequence] polypeptide comprises [about five to] at least about fifteen amino acids.

C4
C14
Claim 46 (Amended). The vaccine of claim 43, [which] further [comprises] comprising an immunologically effective amount of a second polypeptide [sequence of the *Helicobacter*] comprising H. pylori cytotoxin associated immunodominant (CAI) antigen or a fragment thereof, which second polypeptide [sequence]: (i) comprises at least [five] about ten amino acids, (ii) can induce the production of antibodies to *Helicobacter pylori*, and (iii) exhibits [substantially] no functional contribution to toxicity, or a substantially reduced functional contribution to toxicity.

47. The vaccine of claim 46 wherein said sequence comprises at least ten amino acids.

Claim 48 (Amended). The vaccine of claim 46, wherein said [sequence] second polypeptide comprises [about five to] at least about fifteen amino acids.

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Claim 49 (Amended). A method of [preparation of] preparing a prophylactic or therapeutic vaccine [which comprises] comprising bringing into association:

- (1) an immunologically effective amount of a [polypeptide sequence of the *Helicobacter pylori* cytotoxin] H. pylori CT polypeptide, which polypeptide [sequence]: (i) comprises at least [five] about ten amino acids, (ii) can induce the production of antibodies to *Helicobacter pylori*, and (iii) exhibits substantially no [contribution to] toxicity, or substantially reduced toxicity, and
- (2) a pharmaceutically acceptable carrier.

Claim 50 (Amended). The method of claim 49, [which] further [comprises] comprising adding an immunologically effective amount of a second polypeptide [sequence of the *Helicobacter*] comprising H. pylori [cytotoxin associated immunodominant ([CAI]) antigen or fragment thereof [amino acid sequence set forth in SEQ ID NO:5], which second polypeptide [sequence]: (i) comprises at least [five] about ten amino acids, (ii) can induce the production of antibodies to *Helicobacter pylori*, and (iii) exhibits [substantially] no functional contribution to toxicity, or a substantially reduced functional contribution to toxicity.

Claim 52 (New). The method of claim 50, wherein the second polypeptide comprises SEQ ID NO:5, or a fragment thereof, which second polypeptide: (i) comprises at least about ten amino acids, (ii) can induce the production of antibodies to *Helicobacter pylori*, and (iii) exhibits no functional contribution to toxicity, or a substantially reduced functional contribution to toxicity.

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Claim 51 (New). A prophylactic or therapeutic vaccine comprising an immunologically effective amount of a recombinantly produced *H. pylori* CT polypeptide, wherein said recombinantly produced polypeptide exhibits substantially no toxicity, or substantially reduced toxicity, and a pharmaceutically acceptable carrier.